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Brief Report

Lessons Learned from Studies on Tumor Suppression by Microcell-Mediated Chromosome Transfer

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One approach for identifying chromosomes that carry putative tumor-suppressor genes is the introduction of individual, normal human chromosomes to the tumor cells of interest (*1–4*). In order to identify human chromosomes that can suppress or modulate tumor-associated phentotypes of tumor cells, we performed chromosome transfer experiments via microcell fusion into cell lines derived from a variety of tumors: neuroblastoma (SK-N-MC), fibrosarcoma (HT1080), uterine endometrial carcinoma (HHUA), renal cell carcinoma (YCR), choriocarcinoma (CC1), uterine cervical carcinoma (SiHa), rhabdomyosarcoma (A204), Wilms' tumor (SK-NEP-1), and Kirsten sarcoma virus-transformed NIH 3T3 cells (DT). We first isolated mouse A9 cells containing a single, normal human fibroblast-derived chromosome integrated with pSV2*neo* plasmid DNA (*5*). Following fusion of microcells from these A9 cells with tumor cells, we isolated microcell hybrids with the introduction of a specific chromosome and examined their tumorigenicity and *in vitro* properties (*6–11*). The following results were obtained (Table 1).

The introduction of chromosome 11 suppressed tumorigenicity of HT1080, SiHa, A204, and SK-NEP-1, but not of SK-N-MC, YCR, CC1, and DT, indicating the function of the putative suppressor gene(s) on chromosome 11 is effective only in specific tumors.

The tumorigenicity of YCR was modulated by the introduction of chromosome 3p, but not by chromosomes X and 11, supporting a hypothesis that loss and/or mutational inactivation of a gene on 3p may play a crucial role in the development of human renal cell carcinoma.

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Table 1. Summary of results on suppression of tumorigenicity in nude mice and the *in vitro* transformed phenotypes of various tumor cell lines following microcell-mediated transfer of a normal human chromosome.

Tumor cell lines	Transferred chromosomes: Suppressive effect	Transferred chromosomes: No suppressive effect	Reference
Neuroblastoma (SK-N-MC)	1	11	(*4*)
Fibrosarcoma HT1080	1,[a] 11	2, 7, 12	(*9*)
Uterine endometrial carcinoma (HHUA)	1,[a] 6, 9, 11	19	(*8*)
Renal cell carcinoma (YCR)	3	11, X	(*7*)
Choriocarcinoma (CC1)	7[a]	1, 2, 6, 9, 11	(*11*)
Uterine cervical carcinoma (SiHa)	11	12	(*6*)
Rhabdomyosarcoma (A204)	11		(*3*)
Wilms' tumor (SK-NEP-1)	11		(*3*)
Kirsten sarcoma virus-transformed NIH 3T3 cells (DT)	1[a]	11, 12	(*10*)

[a]Various *in vitro* transformed phenotypes were also suppressed.

The introduction of chromosome 1 suppressed the tumorigenicities of SK-N-MC, HT1080, HHUA, and DT, but not of YCR and CC1, indicating that chromosome 1 also carries tumor-suppressor activity for some tumor cells. The tumorigenicity of HHUA was also suppressed by chromosomes 6 and 9. Only the HT1080, HHUA, and DT microcell hybrids with the introduction of chro-

mosome 1 had concomitant alterations in cellular morphology and *in vitro* transformed properties.

Thus, lessons learned from the above results are as follows. Different members of the family of tumor-suppressor genes are present on different chromosomes. More than one normal chromosome suppresses the tumorigenicity of a given tumor cell line, which indicates that multiple tumor-suppressor genes are involved in certain tumors. Functionally distinct tumor-suppressor genes exist. Gene dosage effects can be observed in some cases.

Microcell hybrids of tumor cells suppressed by different normal chromosomes may be useful in mapping and cloning tumor-suppressor genes as well as in elucidating their function in cell growth and differentiation.

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